IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of: Confirmation No. 8747
Applicant: Norihito SHIMONO et al.
Appln. No.: 10/048,063 Group Art Unit: 1615
Filed: January 28, 2002 Examiner: Micah Paul YOUNG
For: SOLID PREPARATION CONTAINING CHITOSAN POWDER AND PROCESS FOR PRODUCING THE SAME

DECLARATION

Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313-1450

Sir:

I, Norihito SHIMONO, a citizen of Japan and residing at No. 3-13, Ibukidainishi-machi 6-chome, Nishi-ku, Kobe-shi, Hyogo, Japan, declare and say as follows.

1. I was graduated from Kyoto University, Faculty of Pharmacy, Department of Pharmaceutical Sciences, Japan in March 1986, and completed the master's course at the same university, Graduated School of Pharmaceutical Sciences in March 1988, and awarded the degree of Doctor of PH from the Kyoto University, Graduated School of Pharmaceutical Sciences in January 2003.

2. Since April 1988 up till the present, I have been an employee of Dainippon Sumitomo Pharma Company, Limited (former Dainippon Pharmaceutical Company, Limited), and I have been engaged in research works of pharmaceutical formulations in Medical Product Research Laboratory of said company.

3. I am one of the inventors of the present U.S. Patent Application No. 10/048,063 and am familiar with the present invention.

4. I have read the cited Lerner et al. USPN 5,840,332 and I know well the contents of the U.S. patent.

5. Under my direction, the following comparative experiments have been done in order to compare the release of drug from the preparations of the present invention and the preparations disclosed in the cited Lerner et al. reference.

(1) Test Preparations

The following preparations were subjected to the experiments.

(1)-1. Preparations (tablets) of Lerner et al. USPN 5,840,332

The Reference Preparations A, B, C, D, and E were prepared along with the formulations disclosed in Examples 3, 4, 5, 7 and 8 of Lerner et al. excepting that the core tablets containing 30 mg sodium salicylate (active ingredient) (each tablet; weighing 300 mg, diameter 10 mm) were prepared by standard direct tableting method using microcrystalline cellulose and calcium pectinate. The core tablets thus obtained (totally 150 g) were coated with a coating suspension of dispersed particles (e.g. calcium pectinate)/water-insoluble polymer (e.g. Eudragit E) so as to give the following reference tablets.

Ref. Prepar. A wherein the coating amount was 75 g.

Ref. Prepar. B wherein the coating amount was 38 g.

Ref. Prepar. C wherein the coating amount was 68 g.

Ref. Prepar. D wherein the coating amount was 113 g.

Ref. Prepar. E wherein the coating amount was 113 g.

The formulations of these preparations are briefly shown in the following Table 1, wherein the main components: the dispersed particles (a) and the water-insoluble polymer (b) as well as the ratios as a solid of the components (a : b) are shown.

(1)-2 Preparations (tablets) of the present invention

The Preparations F, G, H, I and J of the present invention were prepared in the same manner as disclosed in Lerner et al, Example 5 excepting that chitosen was used instead of calcium pectinate and further that the ratios of the dispersed particles (a) (= chitosan) and the water-insoluble polymer (b) (= Eudragit RS) were varied in 4:1, 2:1, 1:1, 1:2, and 1:4, respectively. The preparations thus prepared was coated as follows.

Prepar. F wherein the coating amount was 113 g.

Prepar. G wherein the coating amount was 34 g or 68 g.

Prepar. H wherein the coating amount was 45 g.

Prepar. I wherein the coating amount was 34 g.

Prepar. J wherein the coating amount was 28 g.

The brief formulations of the preparations of the present invention are also shown in Table 1.

(1)-3 Preparations (pellets) of the present invention

The Preparations K and L (in the form of pellets) of the present invention were prepared by the following method.

Commercially available pellets containing acetaminophen (active ingredient) Nonpareil® 103 (purified sucrose spheres; 16/24 mesh, manufactured by Freund Industrial Co., Ltd., 1450 g) were subjected to powder coating of acetaminophen (900 g) using as a binding solution an aqueous solution of hydroxypropyl methylcellose 2910 to give medicament-containing cores. The cores (360 g) thus obtained were coated with a 4 w/w% solution of Eudragit RS in ethanol (2100 g) wherein chitosan (168 g) was dispersed to give pellets (Preparation K).

Said pellets obtained above were further coated with an enteric coating solution [i.e. a 5 w/w% solution of Eudragit L100-55 in ethanol (1500 g) wherein magnesium stearate (80 g) was suspended] in usual manner to give enteric coating pellets (Preparation L).

The brief formulations of these Preparations K and L of the present invention are also shown in Table 1.

Table I

Test Preparation	a: Dispersed particles	b: Water-insoluble polymer	a:b
Ref. Prepar. A (=Ex.3 of Lerner et al.)	Calcium pectinate	Eudragit E	7:3
Ref. Prepar. B (=Ex.4 of Lerner et al.)	Calcium pectinate	Ethyl cellulose	7:3
Ref. Prepar. C (=Ex.5 of Lerner et al.)	Calcium pectinate	Eudragit RS	7:3
Ref. Prepar. D (=Ex.7 of Lerner et al.)	Crospovidone	Eudragit E	7:3
Ref. Prepar. E (=Ex.8 of Lerner et al.)	Microcrystalline cellulose		7:3
Preparations of the present invention: Prepar. F	Chitosan	Eudragit RS	4:1
Prepar. G			2:1
Prepar. H			1:1
Prepar. I			1:2
Prepar. J			1:4
Prepar. K (pellet)			2:1
Prepar. L (pellet)			2:1

(2) Experiments

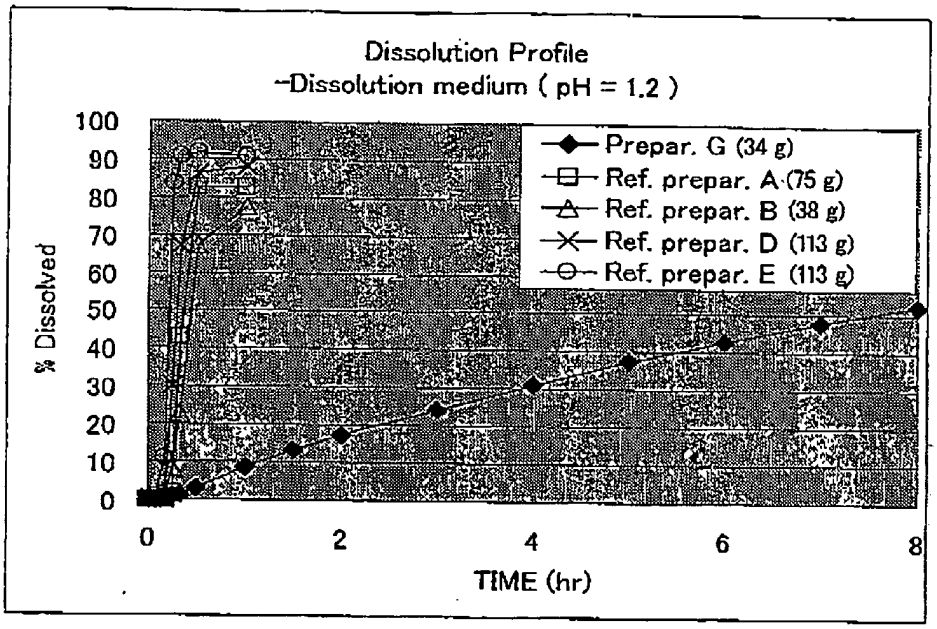
(2)-1 Experiment 1:

Comparison of release of active ingredient with 1st fluid (artificial gastric juice):

The dissolution profile (release of the active ingredient) was tested with respect to the tablet preparations of Lerner et al. (Ref. Prepar. A, B, D, E) and the preparation of the present invention (Prepar. G) by Paddle

method as defined in Japanese Pharmacopeia using 1st fluid (pH 1.2, 900 ml) at 50 rpm, 37°C. The results are shown in the following Fig. 1.

Fig. 1



As is shown in Fig. 1, in the reference preparations (tablets) the active ingredient was dissolved out (released) very rapidly even though these reference preparations had comparatively larger coating amount (75, 38, 113, and 113 g per 115 g of core tablets, respectively) which was larger than the coating amount of the preparation of the present invention (34 g/115 g core tablets). It is assumed that in the reference preparations, the tablets would be swollen with the 1st fluid and then disintegrated, by which the active ingredient was dissolved out very rapidly.

On the other hand, in the preparation of the present invention, the coating of the tablets would not be swollen and the original tablet form was kept for more than 8 hours, and thereby the active ingredient was dissolved out very gradually.

Thus, it is considered that the release of active ingredient with 1st fluid (pH 1.2) will be effected by different mechanism between the reference preparations (the dispersed particles: calcium pectinate, crospovidone, or microcrysalline cellulose) and the preparation of the present invention (the dispersed particles: chitosan), which would have

never been predicted from the cited Lerner et al. reference.

(2)-2 Experiment 2

Comparison of release of active ingredient with changing the dissolution media: 1st fluid (pH 1.2) - 2nd fluid (pH 6.8) - an acidic aqueous solution (pH 4.0):

For simulating the dissolution of drug (release of active ingredient) under the conditions of pH values and passing period of time in digestive tract of biobody in case of oral administration, the test preparations were subjected to the dissolution test by treating with 1st fluid (artificial gastric juice, pH 1.2, for 2 hours), 2nd fluid (artificial intestinal fluid, pH 6.8, for 3 hours) and an acidic aqueous solution (simulated to the conditions in large intestinal, pH 4.0, for 3 hours).

With respect to Ref. Prepar. C of the Lerner et al. and Prepar. G of the present invention, which were prepared by coating (in an amount of 68 g of the coating layer in dry state) on the core tablet (150 g), the dissolution profile was tested by Basket method as defined in Japanese Pharmacopeia (at 25 rpm, 37°C) with dissolution medium of 1st fluid (pH 1.2, 900 ml) for 2 hours, with 2nd fluid (pH 6.8, 900 ml) for 3 hours, and with an acidic aqueous solution (pH 4.0, 900 ml) for 3 hours. The dissolution profile is shown in Fig. 2 hereinafter.

Besides, appearance of the tablets was also observed. The results are shown in Table 2.

Table 2

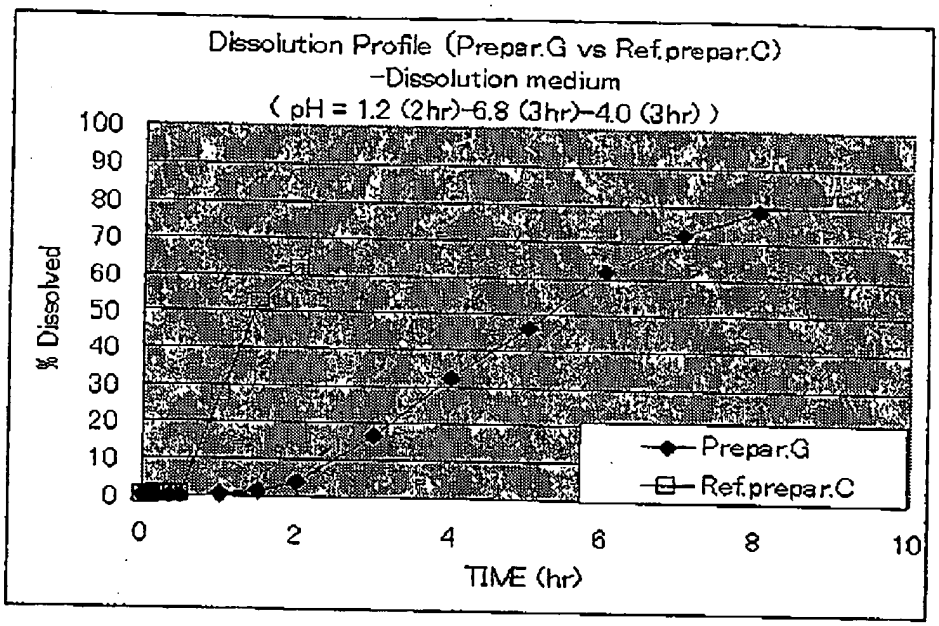
Treating time	0hr	0.5hr	1hr	2hr	5hr	8hr
Prepar. G	o	o	o	o	o	o
Ref. Prepar. C	o	□	×	×	×	×

o : No change □ : Swollen × : Disintegrated

As is shown in Table 2, the Ref. Prepar. C of Lerner et al. was swollen within a very short period of time after initiation of treatment with 1st fluid (pH 1.2) and the coating layer was broken within 1 hour to result in disintegration of the tablets. The same behavior was also observed with respect to Ref. Prepar. B (using ethyl cellulose as the water-insoluble polymer).

Fig. 2 showing the dissolution profile is as follows.

Fig. 2



As is shown in Fig. 2, under the dissolution conditions as mentioned above, the active ingredient was not dissolved out sufficiently from Ref. Prepar. C. It is assumed to be due to the reason that the disintegrated tablets were piled up within the basket and thereby the disintegrated tablets could not sufficiently contact to the dissolution medium. Moreover, the Ref. Prepar. C was disintegrated by contacting with 1st fluid (ph 1.2) and hence could no more subjected to the dissolution test with other dissolution media.

On the other hand, the Prepar. G of the present invention could keep the original shape of tablet through testing for 8 hours, and hence the dissolution test could be completed with all of dissolution mediums. That is, the preparation of the present invention could be subjected to the dissolution test with 1st fluid (pH 1.2) for 2 hours without disintegration, during which the active ingredient was released gradually. Thereafter, the preparation of the present invention could be subjected to the dissolution tests with 2nd fluid (pH 6.8) for 3 hours and further with an acidic aqueous solution of pH 4.0 for 3 hours, during which the active ingredient was released gradually without any disintegration of tablets.

The reference preparation which was coated with a coating suspension of calcium pectinate/Eudragit RS showed entirely different dissolution profile from that of the present invention which was coated with a coating suspension of chitosan/Eudragit RS. Although the same water-insoluble polymer (Eudragit RS) was used in both of Ref. Prepar. C

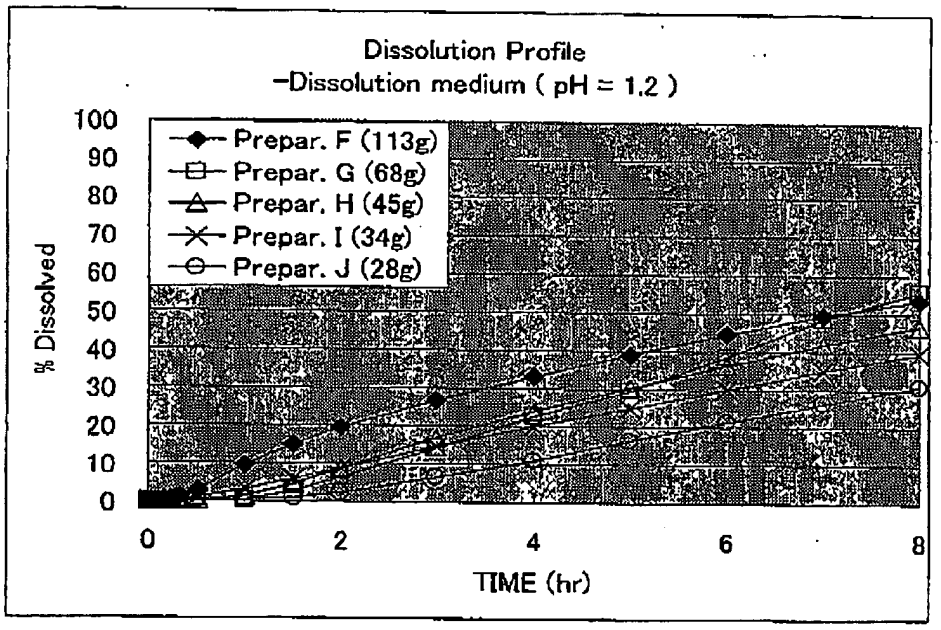
of Lerner et al. and Prepar. G of the present invention, owing to the difference in the dispersed particles, either calcium pectinate or chitosan, the preparations showed entirely different dissolution profile, which would have never been predicted from the cited Lerner et al. reference.

(2)-3. Experiment 3

Comparison of release of active ingredient when varying the ratio of the dispersed particles (a) : the water-insoluble polymer (b):

With respect to Prepar. F, G, H, I, and J having various ratios of the dispersed particles (chitosan) to the water-insoluble polymer (Eudragit RS), the dissolution test with 1st fluid (pH 1.2) was done, and the dissolution profile of these preparations of the present invention was obtained. The results are shown in Fig. 3.

Fig. 3



As is clear from the above Fig. 3, all of the Prepar. F, G, H, I and J of the present invention could show the desired dissolution profile, i.e. sustained release of the active ingredient for a long period of time over 8 hours without disintegration of tablets. When compared these data

with the results in Experiments 1 and 2, these preparations of the present invention showed superior sustained release of the active ingredient in comparison with Ref. Prepar. A, B, C, D, and E of the cited

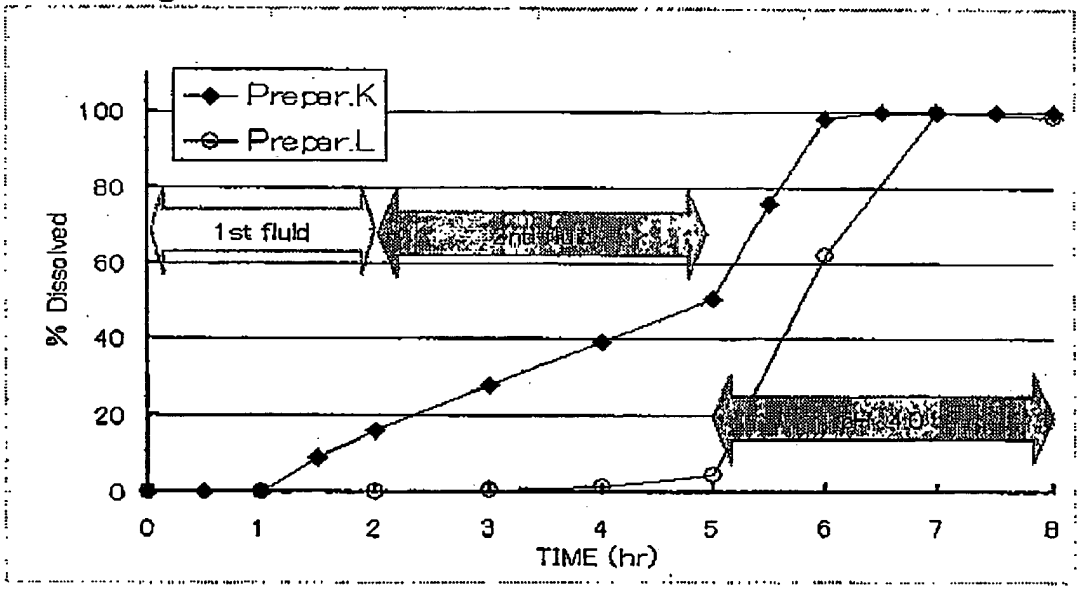
Lerner et al. reference.

(2)-4. Experiment 4

Test of release of active ingredient with changing the dissolution media: 1st fluid (pH 1.2) - 2nd fluid (pH 6.8) - an acidic aqueous solution (pH 4.0):

With respect to the pellet preparations (Prepar. K and L) of the present invention, the dissolution profile was tested with various dissolution media simulated to the digestive tract of biobody in the same manner as described in Experiment 2. The results are shown in the following Fig. 4.

Fig. 4



As is clear from Fig. 4, the sustained release preparation of the present invention (Prepar. K) showed a constant release of the active ingredient by treating with 1st fluid (artificial gastric juice) as well as with 2nd fluid (artificial intestinal fluid). This will be due to that chitosan contained in the coating layer was somewhat dissolved by an acidic solution but was not dissolved by an alkaline solution.

On the other hand, the enteric coating colonic delivery preparation (Prepar. L) of the present invention did not dissolve by treating with 1st fluid (artificial gastric juice, for 2 hours) and with 2nd fluid (artificial intestinal fluid, for 3 hours).

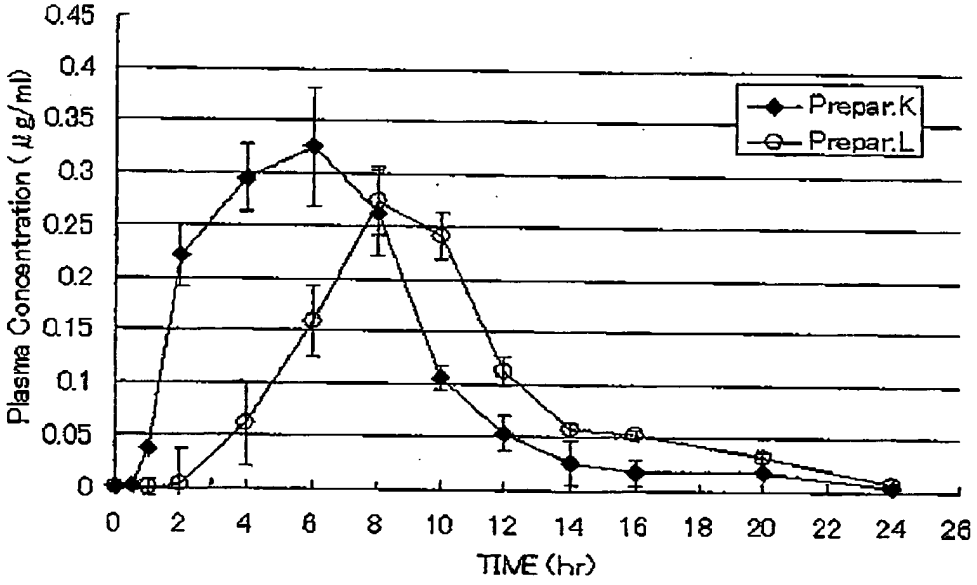
Thus, the release of active ingredient of the preparations of the present invention could be well controlled by regulating the ratio of chitosan to the water-insoluble polymer (Eudragit RS) and the coating amount so that the inner coating layer does not dissolve by the treating with the intestinal fluid (e.g. 2nd fluid, pH 6.8) for 3 hours and release gradually the active ingredient in the large intestine tract (i.e. by treating with an acidic aqueous solution of pH 4.0).

(2)-5. Experiment 5

In vivo test of release of active ingredient in rats:

The pellet preparations (Prepar. K and L) of the present invention were orally administered to rats and the blood concentration of the active ingredient was measured after oral administration. The results are shown in the following Fig. 5.

Fig. 5



As is shown in Fig. 5, the sustained release preparation (Prepar. K) of the present invention showed sustained release property for 12 hours or longer. It was assumed that a rapid release of the active ingredient was observed at the time passing through the stomach in rat (e.g. about 2 hours after administration) and also at the time reaching to the large intestine (e.g. about 5 hours after administration).

On the other hand, the enteric coating colonic delivery preparation (Prepar. L) showed significant extension of Tmax, which suggested that the active ingredient was released at the lower part of the large intestine and absorbed in the biobody.

Thus, it was experimentally confirmed that the dissolution test in Experiment 4 which was simulated to passing through the digestive tract in biobody well corresponded to the *in vivo* test in rats.

It is my opinion based upon my knowledge and experience in this field that the reference preparations disclosed in the cited Lerner et al. reference released the active ingredient very rapidly by the treatment with 1st fluid (artificial gastric juice), which means that it is difficult to control the rapid release of the active ingredient in the stomach and hence is not suitable for sustained release preparation; on the other hand, the preparations of the present invention using chitosan as the main component in the coating layer can give the desired sustained release of the active ingredient from the stomach to the large intestine with keeping the original shapes of tablets/pellets without swelling or disintegration, that is, the preparations of the present invention could show continuous release of the active ingredient by the treatment with 1st fluid (pH 1.2, for 2 hours), 2nd fluid (pH 6.8, for 3 hours) and an acidic aqueous solution (pH 4.0, for 3 hours) which is simulated to the conditions (pH value and time of passing through) in the digestive tract in biobody;

that the preparations disclosed in the cited Lerner et al. reference are rapidly disintegrated by the treatment with 1st fluid (artificial gastric juice), and hence it is assumed that these preparations will be rapidly disintegrated when administered orally, and hence, it will be unable to give sustained release property to these preparations of the Lerner et al. reference, in other words, these preparations of Lerner et al. will be not able to tolerate the shear which will be loaded within the stomach;

that it would never been predicted from the cited Lerner et al. reference that the release of active ingredient within the stomach can be controlled by using chitosan coating like in the present invention, because it is not expected to improve such a release of active ingredient in the stomach by suspending dispersed particles in a water-insoluble polymer for the coating medium as specifically disclosed in the cited Lerner et al. reference;

that as is seen from Experiment 2, under the conditions simulated to the case of oral administration, i.e. by treating with 1st fluid (artificial

gastric juice, pH 1.2, for 2 hours), 2nd fluid (artificial intestinal fluid, pH 6.8, for 3 hours) and an acidic aqueous solution (simulated to the conditions in large intestinal, pH 4.0, for 3 hours), the preparation of Lerner et al. reference (Ref. Prepar. C) and the preparation as claimed in claim 7 of the present invention (Prepar. G) showed significantly different dissolution profile, and from this viewpoint, the present invention would never been predictable from the cited Lerner et al. reference; and

that as is seen from Experiment 4, the enteric coating colonic delivery preparation of the present invention can keep the original shape of preparation (tablets and pellets) even by treating with 1st fluid (pH 1.2, for 2 hours) and 2nd fluid (pH 6.8, for 3 hours), that is, can keep the original shape with releasing the active ingredient in very small amount (i.e. substantially no release of the active ingredient) for about 5 hours after administration and when reached to around the large intestine (after about 4-5 hours) the blood concentration of the active ingredient is increasing, and hence, by applying an enteric coating to the preparation as claimed in claim 7, the resulting enteric coating preparation of the present invention is suitable as a colonic delivery preparation, which would never been predicted from the disclosure of the cited Lerner et al. reference.

The undersigned declares further that all statements made herein of his own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the above-identified application or any patent issuing thereon.

This 25th day of July, 2006

Norihito Shimono

Norihito Shimono